

CLAIMS

1. A method of optimizing secretion of a heterologous polypeptide of interest in a cell comprising comparing the levels of expression of the polypeptide under control of a set of nucleic acid variants of a translation initiation region, wherein the set of variants represents a range of translational strengths, and determining the optimal translational strength for production of mature polypeptide, wherein the optimal translational strength is less than the translational strength of the wild-type translation initiation region.

2. The method of claim 1, wherein the variants comprise nucleic acid variants of a secretion signal sequence.

3. The method of claim 2, wherein the variant secretion signal sequences are variants of STII.

4. The method of claim 3, wherein the STII variants are the following variants

5' TCTAGAGGTTGAGGTGATTTT ATG AAA AAG AAT ATC GCA TTT CTT CTT
5 GCA TCT ATG TTC GTT TTT TCT ATT GCT ACA AAY GCS TAT GCM 3' (SEQ ID NO:15);

5' TCTAGAATT ATG AAA AAG AAT ATC GCA TTT CTT CTT GCA TCT ATG TTC
10 GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:16);

5' TCTAGAATT ATG AAG AAG AAT ATT GCG TTC CTA CTT GCC TCT ATG TTT
GTC TTT TCT ATA GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:17);

5' TCTAGAATT ATG AAG AAG AAT ATC GCA TTT CTT CTT GCA TCT ATG TTC
15 GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:18);

5' TCTAGAATT ATG AAA AAA AAC ATC GCA TTT CTT CTT GCA TCT ATG TTC
GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:19);

5' TCTAGAATT ATG AAA AAA AAC ATT GCC TTT CTT CTT GCA TCT ATG TTC
20 GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:20);

5' TCTAGAATT ATG AAG AAA AAC ATC GCT TTT CTT CTT GCA TCT ATG TTC
25 GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:21);

5' TCTAGAATT ATG AAA AAG AAC ATA GCG TTT CTT CTT GCA TCT ATG TTC
GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:22); and

5' TCTAGAGGTTGAGGTGATTTT ATG AAA AAA AAC ATC GCA TTT CTT CTT
30 GCA TCT ATG TTC GTT TTT TCT ATT GCT ACA AAC GCG TAT GCM 3' (SEQ ID NO:23).

ADD 4)